phosphate backbone and their impact on the folding free energy, we have formulated a mathematical treatment for computing the volume of the main-chain torsion angle conformation space between every pair of nucleobases along any sequence and the corresponding backbone entropy. We compare the computed conformational entropies against a statistical free energy analysis of structures in the crystallographic database from several thousand backbone conformations between nearest-neighbor nucleobases as well as against computer simulations. Using this calculation we have analyzed the backbone entropy of several ribozymes and riboswitches and found that their entropic strains are highly localized. This suggests the folding and stability of the RNA structures are critically dependent on these local entropic strain domains and leave the rest of the sequence relatively flexible. The total entropic penalty in the backbone for the native fold can be as high as 0.7 cal/K/mol per nucleotide for these medium and large RNAs, producing a contribution to the overall free energy of up to 40 kcal/mol for a 200-nucleotide structure. We also look at the correlation of nucleotide conformations along several sample loop sequences to determine the effect of nucleotides beyond the nearest-neighbor.

Membrane Physical Chemistry I

1459-Pos Board B189

Effects of Oxidized Lipid Species on Permeability of Giant Unilamellar Vesicle Membranes

Kristina A. Runas, Su Li, Noah Malmstadt.
University of Southern California, Los Angeles, CA, USA.

Oxidation of unsaturated lipids in cellular membranes has been shown to cause severe membrane damage and potentially cell death. Even in low concentrations, oxidized lipid species are known to cause changes in the membrane structure, such as decreased fluidity. Vesicles containing concentrations of oxidized species as low as 20mol% total lipid concentration can display the spontaneous formation of pores. Below this poration limit, the effects of oxidation on membrane permeability have not been quantified. Here, we use giant unilamellar vesicles (GUVs) as a system to measure passive transport across membranes containing defined concentrations of oxidized lipid species. GUVs consisting of a saturated phospholipid, an unsaturated phospholipid, and cholesterol were used as a model membranes. By replacing defined amounts of the unsaturated lipid with a corresponding oxidized product, the oxidation process could be mimicked, yielding vesicles of varying oxidized lipid concentration. Oxidized lipid concentration was varied from 0mol% to 15mol% of the total lipid concentration. We measured passive transport across the membrane using a microfluidic trap to capture the vesicles and spinning disk confocal microscopy to track the transport of a fluorescent species in the equatorial plane of each GUV analyzed. We used fluorescently labeled short-chain poly(ethylene glycol) species of various molecular weights to track the diffusion of a representative small molecule. Membrane permeability was determined by fitting the resulting concentration profiles to a finite element model of diffusion and permeation around and through the membrane. Experiments showed that an increase in oxidized concentration increases membrane permeability. As passive transport is an important mechanism for drug delivery, understanding the relationship between oxidation and permeation could provide insight into the pharmaceutical characteristics of oxidized cells.

1460-Pos Board B190

The Influence of Hydroxyl Position on Oxysterol/Phospholipid Monolayer Phase Behavior: Experimental Results and Model

Joan C. Kunz[1], Eleni Beyene[1], Luis H.B. Hernandez[2], Ravi Tavakley[2], Benjamin L. Stottrup[2].
[1]Chemistry, Augsburg College, Minneapolis, MN, USA, [2]Physics, Augsburg College, Minneapolis, MN, USA.

Oxysterols are naturally produced by enzymatic and non-enzymatic processes. Increased oxysterol concentration has been correlated with cell age, membrane thinning in model systems, and several pathologies. It has been previously reported that mixed phospholipid monolayers containing 25-hydroxycholesterol exhibit anomalous phase behavior compared to similar cholesterol containing monolayers. We present a systematic series of Langmuir monolayer and fluorescence microscopy studies which focus on the role of the positions of the hydroxyl moieties in difunctional oxysterols. The oxysterols 20, 22, 25, and 27 hydroxycholesterol were selected and mixed with DMPC for compositions from 10 - 90 mole percent oxysterol (increments of 10 percent were chosen). In all of these systems phase behavior was consistent with the 25-hydroxycholsterol/DPPC system. Specifically, an upper and lower miscibility phase transition were observed in all systems for oxysterol concentrations less than 40 mole percent and a discontinuity in the pressure-area isotherm directly tracked the lower miscibility transition pressure. Likewise the area-expansion due to the presence of the oxysterols was confirmed. We present a model for oxysterol behavior within lipid monolayers based on the presence and location of the two hydroxyl groups on the oxysterols. Consistent with our model we find the lower transition pressure increases with increasing distance between the two hydroxyl moieties. Pressure-area isotherms, fluorescence microscopy, analysis of domain size distribution, phase fraction measurements, isobaric cuts, and phase diagrams will be used to support our model of phospholipid/oxysterol interactions.

1461-Pos Board B191

Surfactants, Salt, and pH Alter Nanoparticle-Model Cell Membrane Interactions

Katie Brennan, Luke Cuculis, Shelli L. Frey.
Chemistry Department, Gettysburg College, Gettysburg, PA, USA.

Due to their small size, nanoparticles have the ability to penetrate pulmonary and vascular tissue, and as a result, are classified as potential human carcinogens. On the other hand, nanoparticle insertion into targeted cells can play a key role in drug delivery and gene therapy applications, prompting a need to more thoroughly characterize nanoparticle/membrane interactions. Polystyrene nanoparticles with modifications in surface functionalization and detergent conditions remain monodisperse in a variety of aqueous solutions as measured by dynamic light scattering, but tend to aggregate in phosphate buffered saline at neutral pH potentially due to electrostatic screening effects. Calcein leakage assays with small unilamellar egg phosphatidylcholine vesicles were run to measure their interactions with nanoparticles and determine applicability of previous experiments performed with lipid monolayers at the air-water interface that modeled the outer leaflet of a cell membrane. In pure water, adding surfactant to detergent-free nanoparticle solutions increased the magnitude of membrane permeabilization compared to nanoparticles alone. In a buffered solution, the opposite was true; addition of surfactant decreased the nanoparticle induced membrane permeabilization. To better understand nanoparticle-detergent-membrane interactions, either nanoparticles or detergents were individually introduced to the vesicles, and following a time lapse, the other component was introduced. A model of detergent sequestration by the polystyrene nanoparticles explains these results and provides insight to the mechanism of vesicle leakage caused by surfactant-nanoparticle mixtures.

1462-Pos Board B192

Tracking the Modulation of Membrane Structure in SUVs by DSC - A Comment on Lipid Phase Transition

Chen Shen, Beate M. Klösgen.
Dept. of Phys. Chem. and Pharm. & MEMPHYS, University of Southern Denmark, Odense, Denmark.

Details of the phase transition of DPPC were tracked by DSC (0.05K/min), using a SUV (Φ=100nm) suspension. The main transition was found to comprise three contributions. We attribute this to separate contributions from the chains in both leaflets, and the headgroups. The model was applied to explain the impact of resveratrol on the membrane.

The pretransition is absent, indicating a low cooperativity between the two leaflets of the SUV. The composite main transition peak requests the assumption of at least three sub-peaks. One sub-transition occurs as a broad peak at 313.3K. We attribute it to the headgroups (13.3kJ/mol). The sub-transition of the chains occurs at 314.0K, as a rather sharp peak with a small shoulder. We attribute this appearance to the different bending conditions of the two leaflets in the SUVs, and a resulting difference in the lateral pressure profile as the chains of the outer layer are on average more packed. Their melting thus requires an excess enthalpy (6.3kJ/mol) in addition to the basic consumption for all chains (14.7kJ/mol). The average cost of the whole transition is 31.5kJ/mol.

Resveratrol locates within the headgroup region. This causes a freezing point depression and an even lower cooperativity. Resveratrol also has a condensing effect. During the SUV preparation it can freely distribute, therefore the chain packing difference between the leaflets can be compensated by asymmetric incorporation of the drug. Accordingly, the chain transition is more homogeneous.

Our findings reveal a detailed picture about the phase transition of a lipid SUV membrane. The DSC thermogram reflects both the impact of the headgroups and of the difference between the two leaflets. This bending effect decays as the curvature decreases, and is eventually absent for planar membranes.